

DR. CARNEVALE: Our last but certainly not least speaker today is a new member of the Bureau of Veterinary Medicine. It gives me great pleasure to introduce Bob Livingston. Bob joined BVM with the merger of the Human Food Safety Section from the Bureau of Foods with the BVM about a year ago. He got his B.S. and Ph.D. degrees in Chemistry from the University of Utah. He worked at the National Bureau of Standards before joining the Bureau of Foods in 1973 and then joined BVM as I said in 1982 as part of the transfer. He is presently Director of the Chemistry staff in the Office of Human Food Safety and what Bob's going to bring to you today are some human safety and residue aspects of the dose determination issue.

DR. LIVINGSTON: Thank you, Rich. Human food safety may be last on the program but it's certainly not least of the regulatory responsibilities of the BVM.

HUMAN FOOD SAFETY ASPECTS
OF DOSE DETERMINATION

Robert C. Livingston, Ph.D.

The use of drugs in food-producing animals may result in drug-related residues in meat, milk, and eggs. Therefore, as a part of the premarket approval process required by the Federal Food, Drug, and Cosmetic Act, conditions of use must be established for animal drugs to assure human safety as well as efficacy and safety to the animal species. Residues from all animal drugs are evaluated for human safety under the General Food Safety Clause of the Act. In addition, those drugs that are carcinogens must satisfy the requirement of the DES proviso which states that "no residues of such drug will be found by a method of examination prescribed or approved by the Secretary".

Three areas are of major concern in the evaluation of the human safety of these drug residues: (1) the level and nature of drug-related residues in edible animal products, (2) the demonstration of the safety of the residues using appropriate toxicity tests, and (3) the development of adequate procedures for setting withdrawal periods and assuring that residues do not exceed the safe level in actual use. My presentation today will be limited to the determination of levels of residues in animal-derived foods from the use of drugs or feed additives, procedures for demonstrating safety of those residues, and the role of dose determination in human food safety.

In the near future FDA will be announcing the availability of six guidelines collectively known as "General Principles for Evaluating the Safety of Compounds Used in Food-Producing Animals" that implement the General Food Safety Clause of the Act. The guidelines are listed in Table 1 and are currently being used in the review of sponsor's applications.

The first guideline titled "Guideline for Metabolism Studies and for Identification of Residues for Toxicological Testing" describes the types of studies a sponsor should conduct to determine the level and nature of the drug-related residues in edible tissues resulting from a particular use of a drug. The sponsor should first measure the depletion of total drug-related residues in edible tissues of target animals at various times after the last administration of the compound. Total residues resulting from a drug administered to an animal consist of the parent drug and all compounds derived from it, including metabolites, and residues bound to biological macromolecules. Total residues are typically determined in all edible tissues by dosing animals that are representative of the proposed target population with a radiolabeled drug. Generally, the sponsor performs the depletion study by administering the radiolabeled drug to a sufficient number of previously unmedicated animals to permit serial sacrifice of groups of animals at intervals after the last treatment. If a drug is to be administered to an animal over a period of time, the radiolabeled drug must be given for 5 to 7 days or for a sufficient period to establish steady-state levels in the tissues if a zero withdrawal is requested. From such an experiment, the depletion of total residues in each of the tissues can be observed. If a drug is intended for use in both male and female animals both sexes should be used in the depletion study because males and females of a species can metabolize chemicals at different rates. The dose should be the highest intended treatment level and should simulate proposed conditions of use. The agency recommends that at least twelve animals be dosed and serially sacrificed at zero withdrawal and three additional withdrawal periods. If the sponsor seeks a zero withdrawal, only six animals need be sacrificed, all at zero withdrawal. The edible tissues from each animal at each withdrawal time are analyzed for total radioactivity which is reported in terms of parent drug equivalents.

Structural identification of major metabolites may be necessary depending on the degree of toxicological concern for the parent compound and its potential metabolites. FDA will consider a metabolite to be a major metabolite if either

(a) it is present in an amount greater than 10% of the total residue in an edible tissue at zero withdrawal, or (b) its concentration exceeds 0.1 ppm at zero withdrawal.

Once the total residues in all edible tissues are determined, the sponsor must demonstrate that they are at or below their safe concentrations at zero withdrawal or some withdrawal period that will be reasonably expected to be followed in practice. The safe concentrations are determined by toxicity studies in laboratory animals. Table 2 lists the basic toxicology package normally required to support an NADA: genetic toxicity tests, two 90-day feeding studies, and a two-generation reproduction study. These studies are discussed in the "Guideline for Toxicological Testing" and protocols are presented in a book titled "Toxicological Principles for the Safety Assessment of Direct Food Additives" or more commonly the "Redbook" [1]. The safe concentration is calculated from the highest no-effect level in the most sensitive species tested. The formulas in Table 3 are used to calculate the safe concentration of total residues. The safety factor for the studies in Table 2 is generally 1000 as proposed in the new guidelines. The calculation of safe concentration assumes a 60 kg man and a 1500 g daily diet of solid food. The food factor is a fraction that represents that portion of the solid diet of each edible tissue in each animal species. The food factor for muscle in all species is 1/3, i.e., the daily diet consists of 500 grams of muscle. Organ meats have smaller food factors by amounts ranging from 1/6 to 1/15 depending on the organ and the species.

It is important to note that safe concentrations are calculated in all edible tissues. Conditions of use of the drug must ensure that residues in all edible tissues are below their safe level. Generally, the tissue that is last to deplete to its safe level is designated the target tissue. The target tissue can be used to ensure safe concentrations of residues in all edible tissues.

Previously, FDA had used a safety factor of 2000 for animal drugs supported only with subchronic toxicity studies. This safety factor was adopted from the 1966 National Academy of Sciences negligible tolerance concept for pesticides. The change to a safety factor of 1000 was done to bring the safety evaluation of animal drug residues in harmony with the evaluation of food additives. Since 1966 a cap for allowable safe concentrations in muscle based on subchronic studies has

been 0.1 ppm, usually denoted as a negligible tolerance. Under the new guidelines, the cap will be raised to 1 ppm in the total solid diet of 1500 g or 3 ppm in muscle.

If the total residues at any practical withdrawal period exceeds the calculated safe concentrations, the sponsor may elect to increase the safe concentrations by conducting the chronic studies listed in Table 4. The same procedures for calculating safe concentrations as described above are used except the safety factor for chronic studies is generally 100. In some cases the sponsor will be required to conduct chronic studies to test for carcinogenicity even when the subchronic studies yield an adequate safe concentration. All compounds to be used in food-producing animals are required to go through a decision-tree to determine whether a significant potential for carcinogenicity exists. The decision-tree is presented in the "Guideline for Threshold Assessment". If the compound is determined to be a carcinogen, the safe concentration is determined by utilizing a modified linear-risk assessment.

In summary human food safety does not place any restrictions on the sponsor other than to require studies in the target animal to be done at the highest dose requested for approval. If the residues in any of the edible tissues are above their safe concentrations at zero withdrawal, a withdrawal period will be established to allow the residues to deplete to their safe levels. Some sponsors have elected to conduct residue studies at higher dose levels than label directions specify; this is the sponsor's prerogative as long as it demonstrates that the resulting residue concentrations are safe. This avoids a reevaluation of human food safety when supplemental applications are submitted that have higher dosing regimens. FDA's goal in human food safety is to publish these six guidelines so that the sponsor can make an informed decision on a potential product based on the market place, preliminary studies on the compound, and the safety evaluation process.

REFERENCES

1. Single copies (Catalog No. PB 83170696) are available from National Technical Information Services, 5285 Port Royal Road, Springfield, Virginia 22161.

TABLE 1

HUMAN FOOD SAFETY GUIDELINES

I. Guideline for Metabolism Studies and for Identification of Residues for Toxicological Testing

II. Guideline for Toxicological Testing

III. Guideline for Threshold Assessment

IV. Guideline for Establishing a Tolerance

V. Guideline for Approval of Methods of Analysis for Residues

VI. Guideline for Establishing Withdrawal Periods

TABLE 2

TOXICITY TESTING GENERALLY
NEEDED FOR SPONSORED COMPOUNDS

- Battery of Genetic Toxicity Tests
- 90-Day Feeding Study in a Rodent and in Non-Rodent Mammalian Species
- 2-Generation Reproduction Study in Rats With a Teratology Component
- Other Specialized Testing if Necessary

TABLE 3 - CALCULATION OF SAFE CONCENTRATIONS FOR DRUG RESIDUES

Required Information

No-effect concentration (NEC)

Safety factor (SF)

Acceptable daily intake (ADI)

Food Factor (FF)

Equations

$$(1) \text{ ADI} = \text{NEC} / \text{SF}$$

$$(2) \text{ Safe Concentration} = \frac{\text{ADI} \times 60 \text{ kg}}{\text{FF} \times 1.5 \text{ kg/day}}$$

TABLE 4

- Chronic bioassays in each of two rodent species
- Chronic bioassay (one year) in a non-rodent mammalian species (usually the dog)
- Teratology study in a second species
- Other specialized testing if necessary